10/594908

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SEQUENCE LISTING

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<110> Wang, Xiangbin
     Huang, Hualiang
     Zhao, Baofeng
      Zhao, Qi
     Piao, Jinhua
     Lin, Qing
<120> A GENETIC ENGINEERING RECOMBINANT ANTI-CEA, ANTI-CD3
     AND ANTI-CD28 SINGLE-CHAIN TRI-SPECIFIC ANTIBODY
<130> 11774-006-999 (I040179)
<150> PCT/CN2005/000408
<151> 2005-03-29
<150> CN 200410032158.3
<151> 2004-04-01
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Lys Gly Lys Ala Thr Phe Thr Gly Asp Val Ser Ser Asn Thr Ala Tyr
65
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Met Lys Leu Ser Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
Ala Thr Gly Thr Thr Pro Phe Gly Tyr Trp Gly Gln Gly Thr Leu Val
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Thr Val Ser Ala Thr Ser Thr Pro Ser His Asn Ser His Gln Val Pro
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        115
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Ser Ala Gly Gly Pro Thr Ala Asn Ser Gly Ser Arg Asp Ile Val Leu
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Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly Gln Arg Ala Thr
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Ile Ser Cys Arg Ala Ser Gln Ser Val Ser Thr Ser Ser Tyr Thr Tyr
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Met His Trp Tyr Gln Gln Lys Pro Gly Gln Pro Pro Lys Leu Leu Ile
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Lys Tyr Ala Ser Asn Leu Glu Ser Gly Val Pro Ala Arg Phe Ser Gly
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Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu Glu
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Glu Asp Thr Ala Tyr Tyr Cys Gln His Ser Trp Glu Ile Pro Arg
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Gly Leu Ile Asn Pro Tyr Lys Gly Val Ser Thr Tyr Asn Gln Lys Phe
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Met Glu Leu Leu Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
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Ala Arg Ser Gly Tyr Tyr Gly Asp Ser Asp Trp Tyr Phe Asp Val Trp
            100
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Gly Ala Gly Thr Ser Val Thr Val Ser Ser Thr Ser Gly Gly Gly
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Ser Gly Gly Gly Ser Gly Gly Gly Ser Ser Arg Asp Ile Gln
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Met Thr Gln Thr Thr Ser Ser Leu Ser Ala Ser Leu Gly Asp Arg Val
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Thr Ile Ser Cys Arg Ala Ser Gln Asp Ile Arg Asn Tyr Leu Asn Trp
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Tyr Gln Gln Lys Pro Asp Gly Thr Val Lys Leu Leu Ile Tyr Tyr Thr
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Ser Arg Leu His Ser Gly Val Pro Ser Lys Phe Ser Gly Ser Gly Ser
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Gly Thr Asp Tyr Ser Leu Thr Ile Ser Asn Leu Glu Gln Glu Asp Ile
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                                             220
Ala Thr Tyr Phe Cys Gln Gln Gly Asn Thr Leu Pro Trp Thr Phe Ala
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cagttetete tgaaactgte tteegtagae accgetgtat actattgtge tegtteetat 1980.
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Lys Pro Gly Ala Ser Val Lys Ile Ser Cys Lys Ala Thr Gly Tyr Thr
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Phe Ser Asp Tyr Trp Ile Glu Trp Val Lys Gln Arg Pro Gly His Gly
        35
Leu Glu Trp Ile Gly Glu Ile Leu Pro Gly Ser Gly Arg Thr Asp Tyr
    50
                        55
                                            60
Asn Glu Arg Phe Lys Gly Lys Ala Thr Phe Thr Gly Asp Val Ser Ser
65
                                        75
                    70
Asn Thr Ala Tyr Met Lys Leu Ser Ser Leu Thr Ser Glu Asp Ser Ala
                85
                                    90
Val Tyr Tyr Cys Ala Thr Gly Thr Thr Pro Phe Gly Tyr Trp Gly Gln
                                                    110
            100
                                105
Gly Thr Leu Val Thr Val Ser Ala Thr Ser Thr Pro Ser His Asn Ser
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His Gln Val Pro Ser Ala Gly Gly Pro Thr Ala Asn Ser Gly Ser Arg Asp Ile Val Leu Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly Gln Arg Ala Thr Ile Ser Cys Arg Ala Ser Gln Ser Val Ser Thr Ser Ser Tyr Thr Tyr Met His Trp Tyr Gln Gln Lys Pro Gly Gln Pro Pro Lys Leu Leu Ile Lys Tyr Ala Ser Asn Leu Glu Ser Gly Val Pro Ala Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His Pro Val Glu Glu Asp Thr Ala Tyr Tyr Tyr Cys Gln His Ser Trp Glu Ile Pro Arg Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys Glu Phe Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Ser Thr Glu Val Lys Leu Val Glu Ser Gly Pro Glu Leu Val Lys Pro Gly Ala Ser Met Lys Ile Ser Cys Lys Ala Ser Gly Tyr Ser Phe Thr Gly Tyr Thr Met Asn Trp Val Lys Gln Ser His Gly Lys Asn Leu Glu Trp Met Gly Leu Ile Asn Pro Tyr Lys Gly Val Ser Thr Tyr Asn Gln Lys Phe Lys Asp Lys Ala Thr Leu Thr Val Asp Lys Ser Ser Ser Thr Ala Tyr Met Glu Leu Leu Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys Ala Arg Ser Gly Tyr Tyr Gly Asp Ser Asp Trp Tyr Phe Asp Val Trp Gly Ala Gly Thr Ser Val Thr Val Ser Ser Thr Ser Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Ser Ser Arg Asp Ile Gln Met Thr Gln Thr Thr Ser Ser Leu Ser Ala Ser Leu Gly Asp Arg Val Thr Ile Ser Cys Arg Ala Ser Gln Asp Ile Arg Asn Tyr Leu Asn Trp Tyr Gln Gln Lys Pro Asp Gly Thr Val Lys Leu Leu Ile Tyr Tyr Thr Ser Arg Leu His Ser Gly Val Pro Ser Lys Phe Ser Gly Ser Gly Ser Gly Thr Asp Tyr Ser Leu Thr Ile Ser Asn Leu Glu Gln Glu Asp Ile Ala Thr Tyr Phe Cys Gln Gln Gly Asn Thr Leu Pro Trp Thr Phe Ala Gly Gly Thr Lys Leu Glu Leu Lys Arg Ala Val Asp Phe Gln Asn Ala Leu Leu Val Arg Tyr Thr Lys Lys Val Pro Gln Val Ser Thr Pro Thr Pro Val Glu Val Ser His Met Gln Val Gln Leu Gln Glu Ser Gly Pro Gly Leu Val Lys Pro Ser Gln Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Phe Ser Leu Ser Asp Tyr Gly Val His Trp Val Arg Gln Pro Pro Gly Lys Gly

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Leu Glu Cys Leu Gly Val Ile Trp Gly Gly Gly Thr Asn Tyr Asn Ser
    610
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                                             620
Ala Leu Met Ser Arg Arg Val Thr Ser Ser Asp Asp Thr Ser Lys Asn
625
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                                                             640
                                         635
Gln Phe Ser Leu Lys Leu Ser Ser Val Asp Thr Ala Val Tyr Tyr Cys
                645
                                                         655
                                     650
Ala Arg Ser Tyr Tyr Tyr Ser Met Asp Tyr Trp Gly Gln Gly Thr Leu
                                 665
Val Thr Val Ser Ser Gly Thr Glu Gln Lys Leu Ile Ser Glu Glu Asp
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Leu Asn Gly Ala Ala His His His His His Glu Gln
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gcggtaccgt taccgcgcgg gtacatcata tgtgagacct ctacaggagt tggagttga 59
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<211> 59
<212> DNA
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<210> 13
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tcgg
                                                                    64
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<211> 59
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<211> 30
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<400> 17
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taatacgact cactataggg ga
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                                                                   25
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1
                                                          15
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Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys
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20 25

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            20
                                 25
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<223> SHA linker 2
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Ala Leu Glu Val Asp Glu Thr Tyr Val Pro Lys Glu Phe Asn Ala Glu
                                                          15
                                     10
Thr Phe Thr Phe His Ala Asp Ile
            20
<210> 56
<211> 11
<212> PRT
<213> Artificial Sequence
<220>
<223> cmyc tag
<400> 56
Glu Gln Lys Leu Ile Ser Glu Glu Asp Leu Asn
1
                                     10
<210> 57
<211> 6
<212> PRT
<213> Artificial Sequence
<220>
<223> His tag
<400> 57
His His His His His
<210> 58
<211> 437
```

```
<212> DNA
<213> Artificial Sequence

<220>
<223> nucleotide sequence showing multiple cloning sites (fig 3)

<400> 58
tataccatgg gtctcgagat gtacccgcgc ggtaacacta gtgaattcaa cagcacgtac 60
cgggttgtaa gcgtcctcac cgtactgcac caggactggc tgaatggcaa ggaatacaaa 120
tgcaagagta cttctagaat gtacccgcgc ggtaacgtcg acttccagaa tgcgctgctg 180
gttcgttaca ccaagaaagt accccaagtg tcaactccaa ctcctgtaga ggtctcacat 240
atgatgtacc cgcgcggtaa cggtaccgcg ctggaagttg acgaaaccta cgttccgaaa 300
gaatttaacg cggaaacctt caccttccac gctgacatcc cgcggatggg gctagcgaac 360
aaaaactcat ctcagaagag gatctgaatg gggccgcaca tcatcatcac catcacgagc 420
aataaggatc cgtcgag
```